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# Nitrogen Fixation in Roots of *Ceanothus*

Article by W. John Hayden, Botany Chair

**R**oots are usually out of sight and, therefore, out of mind. But as any good gardener will tell you, it is of utmost importance to understand those unique plant organs, even if their essential functions occur hidden from cursory observation. The red roots of the 2019 VNPS Wildflower of the Year, *Ceanothus americanus*, are particularly important because they host symbiotic bacteria that perform the essential function of nitrogen fixation. These prominent, knobby, distinctively pigmented roots are also the inspiration for the common name Redroot, applied to many species in the genus.

Nitrogen is important. Nitrogen-containing molecules are diverse and absolutely essential for life. It is no exaggeration to say that proteins make up much of the physical structure of cells; proteins are also responsible—as enzymes—for mediating the majority of biochemical reactions that constitute life at the chemical level. Proteins are built from chains of amino acids, and every amino acid includes at least one atom of nitrogen (a few amino acids have two nitrogen atoms). Nitrogen is also present in DNA and RNA, so, through these molecules, nitrogen contributes substantially to the storage, expression, and inheritance of genetic information. Then there is ATP, a relatively small molecule that functions as a little packet of usable energy that fuels all sorts of activity at the molecular level. And let's not forget the hugely diverse category of molecules called alkaloids, synthesized by many different plants, largely to deter herbivory, but also having impact on people as poisons or, beneficially, as medications. It is impossible to imagine life on Earth without

nitrogen-containing compounds.

But there is a paradox about nitrogen and life. You, me, the chickens in my backyard, and the Pawpaw tree that shades them are all constantly bathed in atmospheric nitrogen gas, yet that form of nitrogen is completely unavailable to our cells, or the cells of my chickens, or my Pawpaw. Every breath I take brings a quantity of fresh air into my lungs, 78 percent of which is nitrogen gas, yet despite the importance of nitrogen-containing compounds for life, all that inhaled nitrogen is exhaled, unused and unmodified. For most life forms, nitrogen gas is inert, despite its importance in the biochemistry of life.

Critically, there are exceptions to the picture painted above. A few different kinds of bacteria can take atmospheric nitrogen gas, rip apart the triple bond that holds its two atoms of nitrogen together, and add enough hydrogen to form two molecules of ammonia. This process, known as biological nitrogen fixation, consumes considerable amounts of energy (provided by ATP, referenced briefly above). Also, nitrogen fixation must be performed in the absence of oxygen, because oxygen disables the nitrogenase enzyme responsible for this essential reaction. Unlike the nitrogen gas from which it is produced, ammonia is much more readily used by cells. Some plants can use the ammonia directly to synthesize the nitrogen-containing organic molecules listed above; of course, the bacterial cells can do this too. The important point is that once the relatively inert nitrogen gas has been converted to ammonia, the chemistry of moving that form of nitrogen into proteins, DNA, RNA, ATP, etc., is relatively easy.



Figure 1. New Jersey Tea or Redroot (*Ceanothus americanus*); the bustle of biological activity evident here is possible thanks to the nitrogen-fixing activity of actinomycete bacteria living inside this plant's roots. (Photo by Betty Truax)

This is the importance of symbiotic biological nitrogen fixation; bacteria convert relatively inert nitrogen gas into ammonia, the host plant gobbles up the fixed nitrogen and converts it into diverse nitrogen-containing organic molecules. As we all know, plants feed the rest of the biosphere. Animals get the nitrogen-containing molecules they need by eating plants or other animals that have eaten plants. (The Bible has it right: "All flesh is grass" [Isaiah 40:6].) Finally, as plants and animals die and decompose, microbes recycle their large nitrogen-containing organic molecules into simpler forms that can be taken up by plants—any kind of plant, not just the special ones that host nitrogen-fixing bacteria. There is a bit more complexity to the nitrogen cycle in its entirety: lightning converts some atmospheric nitrogen gas into oxidized forms that plants can assimilate, and there are other types of bacteria that take oxidized forms of nitrogen and convert it back to relatively inert

(See *Ceanothus*, next page)

## Botanical art exhibit reflects journey across America

The Botanical Art Society of the National Capital Region cordially invites members of the Virginia Native Plant Society to the opening reception of “All in a Garden Green” at Green Spring Gardens Park on Sunday, September 8, from 1-3 p.m.

The exhibit is inspired by English writer Frances Trollope who, in 1828, began a four-year journey around the United States. In her book, *Domestic Manners of the Americans* (1832), she noted with delight the plants she encountered. Trollope spent most of 1830

in houses in Washington, Potomac, and Alexandria. Green Spring is an ideal venue to display pictures of the kinds of plants that she might have encountered on her American adventure. Subjects will include plants native to the eastern U.S., as well as those that were cultivated on early American estates such as Green Spring.

The exhibit will be held at the Horticultural Center (4603 Green Spring Rd., Alexandria, VA 22312) and runs from August 27 to October 27.

—Elena Maza Borkland



Illustrations such as this Flame Azalea by Elena Maza Borkland will be on display at Green Spring Gardens until late October as part of the “All in a Garden Green” exhibit sponsored by the Botanical Art Society of the National Capital Region.

## Ceanothus continued

atmospheric nitrogen gas. The point is that nitrogen fixation and subsequent recycling of fixed nitrogen accounts for the majority of the ways in which nitrogen sustains life on this good Earth.

The most well known examples of symbiosis-based biological nitrogen fixation involve *Rhizobium* and closely related bacteria that inhabit the root nodules of legumes. In essence, the legume plant feeds the bacterium energy-rich molecules derived from photosynthesis from which the *Rhizobium* bacteria generate ATP that fuels the production of ammonia from nitrogen gas; a portion of the ammonia produced is released to the plant for its diverse biochemical needs. Vast amounts of nitrogen enter the biosphere by means of legumes and their nitrogen-fixing bacterial symbionts.

But the legume–*Rhizobium* partnership is not the only nitrogen-fixing symbiotic system. Cycads have special roots that grow near the soil surface, and, internally, these roots harbor cyanobacteria (blue-green algae) that perform nitrogen fixation. The peculiar floating aquatic fern *Azolla* hosts cyanobacteria inside

its leaves that serve the same purpose. And free-living (i.e., nonsymbiotic) cyanobacteria also perform nitrogen fixation.

The roster of bacteria that can perform symbiotic nitrogen fixation includes one additional group known as actinomycetes. These are minute, filamentous, bacteria that, decades ago, were thought to be fungi, hence the “-mycetes” suffix in their name. When present symbiotically, actinomycete bacteria inhabit plant roots, which alters their morphology; generally, actinomycete-inhabited roots are enlarged and knobby, compared with uninfected roots. Host plants of nitrogen-fixing actinomycetes are diverse, including Alders (*Alnus.*), Sweetfern (*Comptonia*), Wax Myrtle (*Morella*, *Myrica*), She-oaks (*Casuarina*), Oleasters (*Elaeagnus*), and Redroots (*Ceanothus*).

In *Ceanothus*, the symbiotic actinomycete has been identified as *Frankia ceanothi*, colonies of which develop inside cortex cells of its nodulated (enlarged, knobby) roots.

Compared with intensively studied legume crops, there is little published material on nitrogen fixation in *Ceanothus*; the few studies that have been published focus on species from the western United States, where *Ceanothus* is diverse and, in at least some plant communities, abundant. Our local Redroot, *Ceanothus americanus*, shares with its western relatives a tendency to inhabit nutrient-poor, arid, rocky soils—the sort of environment in which having a built-in nitrogen fertilizer factory would provide an obvious adaptive advantage.

Next time you encounter a specimen of our 2019 Wildflower of the Year and admire its handsome foliage, its intricate small flowers, and the diverse pollinators attracted thereto (Figure 1), take a moment to acknowledge also the hardworking actinomycete cells, sight unseen, collaborating with *Ceanothus* root cells in the task of converting atmospheric nitrogen into the nitrogen-containing organic molecules essential to all the life forms that surround us ... and that, indeed, are essential to us too. ❖